extension or amplification reaction. Such molecules are useful as hybridization probes and in nucleic acid sequencing.

In detail, the invention as described in application serial No. 08/005,061 provides a method for generating a desired single-stranded nucleic acid molecule having a sequence complementary to that of a target nucleic acid molecule, the method comprising the steps:

a) incubating the target molecule in the presence of a primer molecule; wherein the primer molecule is capable of hybridizing to the target molecule, and wherein the primer molecule contains a nucleotide that is substantially incapable of being eliminated by a 5' 3' exonuclease;

b) permitting template-dependent extension of the primer molecule to thereby form the desired nucleic acid molecule; and

c) incubating the target molecule in the presence of a 5' 3' exonuclease, wherein the incubation results in the elimination of the target molecule, and thereby generates the desired single-stranded molecule.

The invention as described in application serial No. 08/005,061 additionally concerns the embodiment of the above method wherein step B additionally includes the substep of incubating the desired nucleic acid molecule in the presence of a second primer molecule capable of hybridizing thereto, and of being extended in a template-dependent manner to thereby form a nucleic acid molecule having a sequence substantially complementary to that of the desired molecule.

The invention as described in application serial No. 08/005,061 additionally concerns the embodiment of the above method wherein step B additionally includes the further substeps of hybridizing the nucleic acid molecule of step B, substep (1), with a complementary primer molecule; and permitting template-dependent extension of the complementary primer molecule to form a nucleic acid molecule having a sequence substantially complementary to that of the target molecule.

The invention as described in application serial No. 08/005,061 additionally concerns the embodiment of the above methods wherein the primer, the phosphorothioate nucleotide derivative, or the single-stranded molecule, or its amplification product is detectably labeled, as with an enzyme label, a fluorescent label, a radioisotopic label, and a chemiluminescent label.

The invention as described in application serial No. 08/005,061 additionally concerns the embodiment of the above methods wherein the desired single-stranded nucleic acid molecule or amplification products are detectably labeled by the incorporation of labeled nucleotides during the template-dependent extension of the primer.

The invention as described in application serial No. 08/005,061 is capable of generating single-stranded molecules regardless of the nature, origin or sequence of the target molecule. Thus, the invention as described in application serial No. 08/005,061 can be used to generate single-stranded molecules that have a naturally occurring sequences, such as a sequence present in a virus (e.g. rhinovirus, hepatitis virus, herpes virus, HIV, etc.), a bacterium (e.g. Escherichia, Clostridium, Mycobacterium, Neisseria, Mycoplasma, Vibrio, Chlamydia, Rickettsia, etc.), a yeast, a fungus, or other lower eukaryote. In particular, the present invention can be used to generate single-stranded molecules that have sequence present in a plant cell, or an animal cell (especially a mammalian cell, such as from a horse, cow, dog, cat or human). The invention as described in application serial No. 08/005,061 can also be used to generate single-stranded molecules that are purely or partially synthetic (i.e. non-naturally occurring). --

I 1 cont'd

At page 51, line 14-17, please delete the phrase "First, the amplification may be mediated using primers that contain 4 posphorothioate-nucleotide derivatives, as taught by Nikiforov, T. (U.S. patent application serial no. 08/005,061)." and replace it by --First, the amplification may be mediated using primers that contain 4 phosphorothioate-nucleotide derivatives.--

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